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EXAMINER	
MUMMERT, STEPHANIE KANE	
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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/700,304	YU ET AL.	
	Examiner	Art Unit	
	Stephanie K. Mummert, Ph.D.	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☐ Responsive to communication(s) filed on _____.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) 1-35 is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) 1-35 is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449 or PTO/SB/08) Paper No(s)/Mail Date 4/12/04.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: _____.

DETAILED ACTION

Claim Rejections - 35 USC § 102

1. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.
>
> (e) the invention was described in (1) an application for patent, published under section 122(b), by another filed in the United States before the invention by the applicant for patent or (2) a patent granted on an application for patent by another filed in the United States before the invention by the applicant for patent, except that an international application filed under the treaty defined in section 351(a) shall have the effects for purposes of this subsection of an application filed in the United States only if the international application designated the United States and was published under Article 21(2) of such treaty in the English language.

2. Claims 1, 4-5, 8-16, 19-21, 24-33 and 35 are rejected under 35 U.S.C. 102(e) as being anticipated by Andersen et al. (US PgPub 2004/0175733; September 2004; effective filing date December 4, 2002). Andersen teaches methods of multiplex amplification of template nucleic acids for gene expression analysis (Abstract).

With regard to claim 1, Andersen teaches a method of producing a sub-population of labeled nucleic acids, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (p. 4, paragraph 34, where first strand cDNA may be synthesized prior to amplification using random RT-primers (e.g., random hexamers or oligo dT primers; p. 4, paragraph 35, where the target polynucleotide may be obtained from virtually any sample or source; p. 5, paragraph 37);

(b) contacting said first strand cDNA with a pool of a representational number of at least 15 distinct gene specific primers under conditions that allow formation of hybrid duplexes between said gene specific primer and said first strand cDNA, wherein each constituent gene specific.

primer has a sequence complementary to a distinct first strand cDNA (p. 4, paragraph 34, where first strand cDNA may be synthesized prior to amplification using random RT-primers (e.g., random hexamers or oligo dT primers and wherein a plurality of PCR primer pairs, at least 100, 300, 500, 1000, and more are used to amplify the target(s) through multiplex amplification; see also Example 1 and 5, where different levels of multiplex amplification are carried out); and

(c) enzymatically extending said gene specific primers from said hybrid duplexes to generate a sub-population of labeled nucleic acids (p. 4, paragraph 34, at least 100, 300, 500, 1000, and more are used to amplify the target(s) through multiplex amplification; see also Example 1 and 5, where different levels of multiplex amplification are carried out; see also p. 6, paragraph 47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable).

With regard to claim 16, Andersen teaches a method of producing a sub-population of labeled nucleic acids, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (p. 4, paragraph 34, where first strand cDNA may be synthesized prior to amplification using random RT-primers (e.g., random hexamers or oligo dT primers; p. 4, paragraph 35, where the target polynucleotide may be obtained from virtually any sample or source; p. 5, paragraph 37);

(b) generating a sub-population of labeled nucleic acids using polymerase chain reaction (PCR) with a pool of a representational number of at least 15 pairs of distinct gene specific primers, wherein one gene specific primer in each pair comprises a sequence complementary to the sense sequence of said distinct gene, and the other primer in the pair comprises a sequence

complementary to the antisense sequence of said distinct gene (p. 4, paragraph 34, where at least 100, 300, 500, 1000, and more are used to amplify the target(s) through multiplex amplification; see also Example 1 and 5, where different levels of multiplex amplification are carried out; see also p. 6, paragraph 47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable; see also p. 5, paragraph 41, where it is noted that each primer pair includes two primers, one forward, one reverse).

With regard to claim 4 and 20, Andersen teaches an embodiment of claims 1 or 16, wherein said sample of RNA comprises total RNA (p. 1, paragraph 10, where it is noted that both DNA or RNA targets may be multiplex amplified; p. 4, paragraph 29; p. 5, paragraph 36).

With regard to claim 5 and 21, Andersen teaches an embodiment of claims 1 or 16, wherein said sample of RNA comprises mRNA (p. 5, paragraph 36, where the target polynucleotide may be RNA (e.g., mRNA, rRNA) and wherein total RNA would inherently comprise mRNA).

With regard to claim 8-9 and 24-25, Andersen teaches an embodiment of claims 1 or 16, wherein said first strand cDNA is synthesized through addition of synthetic random primers or oligo dT primers (p. 4, paragraph 34, where first strand cDNA may be synthesized prior to amplification using random RT-primers (e.g., random hexamers or oligo dT primers).

With regard to claim 10-11 and 26-27, Andersen teaches an embodiment of claims 1 or 16, wherein said pool of gene specific primers comprises at least 20 or at least 50 distinct gene specific primers (p. 4, paragraph 34, where the number of primer pairs may be at least 100, 300, 500, 1000, 10000 or 30000; see also Example 1, where a 95-plex amplification was carried out and Example 5, where higher multiplex amplifications were carried out).

With regard to claim 12, Andersen teaches an embodiment of claim 1, wherein said pool of gene specific primers comprises one oligonucleotide sequence for a single gene (p. 14, Example 1, paragraph 104, where each 20x gene expression product contained two unlabeled amplification primers and one FAM-labeled TaqMan probe, wherein this mixture comprises one sequence for one gene).

With regard to claim 13, Andersen teaches an embodiment of claim 1, wherein said pool of gene specific primers comprises more than one oligonucleotide sequences for a single gene (p. 14, Example 1, paragraph 104, where each 20x gene expression product contained two unlabeled amplification primers and one FAM-labeled TaqMan probe, wherein this mixture comprises more than one sequence for one gene).

With regard to claim 14 and 28, Andersen teaches an embodiment of claims 1 and 16, wherein the said label is directly detectable (p. 6, paragraph 47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable).

With regard to claim 15 and 29, Andersen teaches an embodiment of claims 1 and 16, wherein the said label is detectable after a subsequent chemical or enzymatic reaction (p. 6, paragraph 47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable, including spectroscopic, photochemical, biochemical, immunochemical, enzymatic or chemical means).

With regard to claim 19, Andersen teaches an embodiment of claim 16, wherein said PCR is performed in multiple cycles (p. 8, paragraph 64 where conventional PCR and RT-PCR

are typically carried out with 0.05U polymerase and for the 95-plex amplification, 10 cycles are carried out).

With regard to claim 30, Andersen teaches a method of analyzing the differences in the expression pattern of the genes of special interest between a plurality of different physiological samples, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (p. 4, paragraph 34, where first strand cDNA may be synthesized prior to amplification using random RT-primers (e.g., random hexamers or oligo dT primers; p. 4, paragraph 35, where the target polynucleotide may be obtained from virtually any sample or source; p. 5, paragraph 37);

(b) contacting a pool of a representational number of at least 15 distinct gene specific primers with said first strand cDNA under conditions that allow formation of hybrid duplexes between said gene specific primers and said first strand cDNA, wherein each constituent gene specific primer has a sequence complementary to a distinct first strand cDNA (p. 4, paragraph 34, where at least 100, 300, 500, 1000, and more are used to amplify the target(s) through multiplex amplification; see also Example 1 and 5, where different levels of multiplex amplification are carried out; see also p. 6, paragraph 47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable); and

(c) enzymatically extending said gene specific primers from said hybrid duplexes to generate a sub-population of labeled nucleic acids (p. 4, paragraph 34, where at least 100, 300, 500, 1000, and more are used to amplify the target(s) through multiplex amplification; see also Example 1 and 5, where different levels of multiplex amplification are carried out; see also p. 6, paragraph

47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable) and

(d) comparing the populations of labeled nucleic acids from each physiological source to identify the differences in the populations (p. 11, paragraph 80-81, where the detected signal is a hybridization intensity and where comparison between different samples would be necessary to establish differential gene expression).

With regard to claim 31, Andersen teaches an embodiment of claim 30, wherein the comparing step comprises:

hybridizing the labeled nucleic acids from each of the distinct physiological samples to an array of nucleic acids stably associated with the surface of a substrate (p. 11, paragraph 80-81, where the product of the multiplex amplification described previously can be applied to solid supports comprising polynucleotides designed for analysis of differentially expressed genes and where the detected signal is a hybridization intensity);

washing off the unbound labeled nucleic acids from the surface to produce a detectable hyblidization patterns for each of the distinct physiological samples (p. 11, paragraph 80-81, where the product of the multiplex amplification described previously can be applied to solid supports comprising polynucleotides designed for analysis of differentially expressed genes and where the detected signal is a hybridization intensity); and

comparing the hybridization patterns for each of the distinct physiological samples (p. 11, paragraph 80-81, where the product of the multiplex amplification described previously can be applied to solid supports comprising polynucleotides designed for analysis of differentially expressed genes and where the detected signal is a hybridization intensity).

With regard to claim 32, Andersen teaches a method of analyzing the differences in the expression pattern of the genes of special interest between a plurality of different physiological samples, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (p. 4, paragraph 34, where first strand cDNA may be synthesized prior to amplification using random RT-primers (e.g., random hexamers or oligo dT primers; p. 4, paragraph 35, where the target polynucleotide may be obtained from virtually any sample or source; p. 5, paragraph 37);

(b) generating a sub-population of labeled nucleic acids using polymerase chain reaction (PCR) with a pool of a representational number of at least 15 pairs of distinct gene specific primers, wherein one gene specific primer in each pair comprises a sequence complementary to the sense sequence of said distinct gene, and the other primer in the pair comprises a sequence complementary to the antisense sequence (p. 4, paragraph 34, where at least 100, 300, 500, 1000, and more are used to amplify the target(s) through multiplex amplification; see also Example 1 and 5, where different levels of multiplex amplification are carried out; see also p. 6, paragraph 47, where it is noted that one or more of the primers can include a label and said label refers to any moiety that can render the nucleotides detectable; see also p. 5, paragraph 41, where it is noted that each primer pair includes two primers, one forward, one reverse); and

(c) comparing the populations of labeled nucleic acids from each physiological source to identify the differences in the populations (p. 11, paragraph 80-81, where the product of the multiplex amplification described previously can be applied to solid supports comprising polynucleotides

designed for analysis of differentially expressed genes and where the detected signal is a hybridization intensity).

With regard to claim 33, Andersen teaches an embodiment of claim 32, wherein the comparing step comprises:

hybridizing the labeled nucleic acids from each of the distinct physiological samples to an array of nucleic acids stably associated with the surface of a substrate (p. 11, paragraph 80-81, where the product of the multiplex amplification described previously can be applied to solid supports comprising polynucleotides designed for analysis of differentially expressed genes and where the detected signal is a hybridization intensity);

washing off the unbound labeled nucleic acids from the surface to produce a detectable hybridization patterns for each of the distinct physiological samples (p. 11, paragraph 80-81, where the product of the multiplex amplification described previously can be applied to solid supports comprising polynucleotides designed for analysis of differentially expressed genes and where the detected signal is a hybridization intensity); and

comparing the hybridization patterns for each of the distinct physiological samples (p. 11, paragraph 80-81, where the detected signal is a hybridization intensity and where comparison between different samples would be necessary to establish differential gene expression).

With regard to claim 35, Andersen teaches an embodiment of claim 32, wherein said PCR is performed in multiple cycles (p. 8, paragraph 64 where conventional PCR and RT-PCR are typically carried out with 0.05U polymerase and for the 95-plex amplification, 10 cycles are carried out).

3. Claims 1, 4-5, 8-14, 16, 19-21, 24-28, 30-33 and 35 are rejected under 35 U.S.C. 102(e) as being anticipated by Wiley et al. (US PgPub 2003/0186246; October 2003). Wiley teaches a method for direct comparison of gene expression values between samples using reverse-transcription-polymerase chain reaction (Abstract).

With regard to claim 1, Wiley teaches a method of producing a sub-population of labeled nucleic acids, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (Example 1, p. 9, paragraph 101, where total RNA was isolated and reverse transcribed using M-MLV reverse transcriptase and oligo dT primer);

(b) contacting said first strand cDNA with a pool of a representational number of at least 15 distinct gene specific primers under conditions that allow formation of hybrid duplexes between said gene specific primer and said first strand cDNA, wherein each constituent gene specific. primer has a sequence complementary to a distinct first strand cDNA (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of amplification occurs where cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out); and

(c) enzymatically extending said gene specific primers from said hybrid duplexes to generate a sub-population of labeled nucleic acids (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of amplification occurs where cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out; p. 4, paragraph 39, where another embodiment includes

fluorescently labeling the sample through labeling the primers/oligonucleotide and/or labeling the dNTPs in the 'extension' reaction).

With regard to claim 16, Wiley teaches a method of producing a sub-population of labeled nucleic acids, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (Example 1, p. 9, paragraph 101, where total RNA was isolated and reverse transcribed using M-MLV reverse transcriptase and oligo dT primer; p. 3, paragraph 21, where it is noted that in order to correlate gene expression patterns with clinical phenotypes, multiple genes may need to be evaluated and multiple types of samples are described at paragraph 22);

(b) generating a sub-population of labeled nucleic acids using polymerase chain reaction (PCR) with a pool of a representational number of at least 15 pairs of distinct gene specific primers, wherein one gene specific primer in each pair comprises a sequence complementary to the sense sequence of said distinct gene, and the other primer in the pair comprises a sequence complementary to the antisense sequence of said distinct gene (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of amplification occurs where cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out).

With regard to claim 4 and 20, Wiley teaches an embodiment of claims 1 or 16, wherein said sample of RNA comprises total RNA (p. 9, example 1, where total RNA was isolated from cells grown in monolayer).

With regard to claim 5 and 21, Wiley teaches an embodiment of claims 1 or 16, wherein said sample of RNA comprises mRNA (p. 9, example 1, where total RNA was isolated from cells grown in monolayer, and wherein total RNA would inherently comprise mRNA).

With regard to claim 8-9 and 24-25, Wiley teaches an embodiment of claims 1 or 16, wherein said first strand cDNA is synthesized through addition of synthetic random primers or oligo dT primers (Example 1, p. 9, paragraph 101, where total RNA was isolated and reverse transcribed using M-MLV reverse transcriptase and oligo dT primer).

With regard to claim 10-11 and 26-27, Wiley teaches an embodiment of claims 1 or 16, wherein said pool of gene specific primers comprises at least 20 or at least 50 distinct gene specific primers (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of amplification occurs where cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out).

With regard to claim 12, Wiley teaches an embodiment of claim 1, wherein said pool of gene specific primers comprises one oligonucleotide sequence for a single gene (Table 2, where for certain genes, a single gene specific primer is included in the method, e.g., IVL, JUNB, TP53).

With regard to claim 13, Wiley teaches an embodiment of claim 1, wherein said pool of gene specific primers comprises more than one oligonucleotide sequences for a single gene (Table 2, where for certain genes, more than one sequence was incorporated for examination of a particular gene sequence, e.g., GSTP1, SULTI).

With regard to claim 14 and 28, Wiley teaches an embodiment of claims 1 and 16, wherein the said label is directly detectable (p. 4, paragraph 39, where another embodiment includes fluorescently labeling the sample through labeling the primers/oligonucleotide and/or labeling the dNTPs in the 'extension' reaction, where a fluorescent label is directly detectable).

With regard to claim 19, Wiley teaches an embodiment of claim 16, wherein said PCR is performed in multiple cycles (p. 3, paragraph 34, where the desired number of amplification cycles is between about 3 to 40 and those in between).

With regard to claim 30, Wiley teaches a method of analyzing the differences in the expression pattern of the genes of special interest between a plurality of different physiological samples, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (Example 1, p. 9, paragraph 101, where total RNA was isolated and reverse transcribed using M-MLV reverse transcriptase and oligo dT primer; p. 3, paragraph 21, where it is noted that in order to correlate gene expression patterns with clinical phenotypes, multiple genes may need to be evaluated and multiple types of samples are described at paragraph 22);

(b) contacting a pool of a representational number of at least 15 distinct gene specific primers with said first strand cDNA under conditions that allow formation of hybrid duplexes between said gene specific primers and said first strand cDNA, wherein each constituent gene specific primer has a sequence complementary to a distinct first strand cDNA (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of

amplification occurs where cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out); and

(c) enzymatically extending said gene specific primers from said hybrid duplexes to generate a sub-population of labeled nucleic acids (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of amplification occurs where cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out) and

(d) comparing the populations of labeled nucleic acids from each physiological source to identify the differences in the populations (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR and wherein the expression of the target genes are quantified by comparing the fluorescent intensities of the spots in the array).

With regard to claim 31, Wiley teaches an embodiment of claim 30, wherein the comparing step comprises:

hybridizing the labeled nucleic acids from each of the distinct physiological samples to an array of nucleic acids stably associated with the surface of a substrate (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR);

washing off the unbound labeled nucleic acids from the surface to produce a detectable hyblidization patterns for each of the distinct physiological samples (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following

quantitative RT-PCR and wherein the expression of the target genes are quantified by comparing the fluorescent intensities of the spots in the array); and

comparing the hybridization patterns for each of the distinct physiological samples (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR and wherein the expression of the target genes are quantified by comparing the fluorescent intensities of the spots in the array).

With regard to claim 32, Wiley teaches a method of analyzing the differences in the expression pattern of the genes of special interest between a plurality of different physiological samples, said method comprising:

(a) synthesizing first strand cDNA from a sample of RNA through reverse transcription, wherein the sample of RNA is obtained from a physiological source (Example 1, p. 9, paragraph 101, where total RNA was isolated and reverse transcribed using M-MLV reverse transcriptase and oligo dT primer; p. 3, paragraph 21, where it is noted that in order to correlate gene expression patterns with clinical phenotypes, multiple genes may need to be evaluated and multiple types of samples are described at paragraph 22);

(b) generating a sub-population of labeled nucleic acids using polymerase chain reaction (PCR) with a pool of a representational number of at least 15 pairs of distinct gene specific primers, wherein one gene specific primer in each pair comprises a sequence complementary to the sense sequence of said distinct gene, and the other primer in the pair comprises a sequence complementary to the antisense sequence (p. 9, Example 1, paragraph 105, where one embodiment of the method is described wherein the first round of amplification occurs where

cDNA is incubated with 9 pairs of gene specific primers; see also p. 9, paragraph 108-109, where 96-multiplex amplification was carried out); and

(c) comparing the populations of labeled nucleic acids from each physiological source to identify the differences in the populations (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR and wherein the expression of the target genes are quantified by comparing the fluorescent intensities of the spots in the array).

With regard to claim 33, Wiley teaches an embodiment of claim 32, wherein the comparing step comprises:

hybridizing the labeled nucleic acids from each of the distinct physiological samples to an array of nucleic acids stably associated with the surface of a substrate (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR);

washing off the unbound labeled nucleic acids from the surface to produce a detectable hybridization patterns for each of the distinct physiological samples (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR and wherein the expression of the target genes are quantified by comparing the fluorescent intensities of the spots in the array); and

comparing the hybridization patterns for each of the distinct physiological samples (p. 4, paragraph 39-40, where in certain embodiments, high density arrays are used to measure PCR products following quantitative RT-PCR and wherein the expression of the target genes are quantified by comparing the fluorescent intensities of the spots in the array).

With regard to claim 35, Wiley teaches an embodiment of claim 32, wherein said PCR is performed in multiple cycles (p. 3, paragraph 34, where the desired number of amplification cycles is between about 3 to 40 and those in between).

Claim Rejections - 35 USC § 103

4. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

5. Claims 2-3, 17-18 and 34 are rejected under 35 U.S.C. 103(a) as being unpatentable over Andersen as applied to claims 1, 4-5, 8-16, 19-21, 24-33 and 35 above, and further in view of Chee et al. (US PgPub 2004/0121364; June 2004). Andersen teaches methods of multiplex amplification of template nucleic acids for gene expression analysis (Abstract).

Andersen teaches the limitations of claims 1, 4-5, 8-16, 19-21, 24-33 and 35 as recited in the 102 rejection stated above. Regarding claims 2-3, Andersen does not teach that the labeled nucleic acids are extended in a unidirectional manner. Regarding claims 17-18, Andersen does not teach that the method of amplification is asymmetric. Regarding claim 34, Andersen does not teach that PCR is performed in one cycle.

With regard to claim 2-3, Chee teaches an embodiment of claim 1, wherein said sub-population of labeled nucleic acids extended from said gene specific primers is generated

through a single or multiple cycles of unidirectional DNA polymerization (p. 35, paragraph 409, where linear amplification can be performed through unidirectional amplification).

With regard to claim 17 and 34, Chee teaches an embodiment of claim 16 and 32, wherein said PCR is an asymmetric PCR (p. 24, paragraph 281, where one primer is present in excess concentration).

With regard to claim 18, Chee teaches an embodiment of claim 16, wherein said PCR is performed in one cycle (p. 23, paragraph 273, where the number of cycles is from 1 to thousands, with 10-100 or 20-50 cycles being preferred).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to have applied the method of amplification taught by Chee to the method of multiplex amplification taught by Andersen. As taught by Chee, "Unidirectional amplification can be performed by priming and polymerase directed extension from a single strand" (p. 35, paragraph 409). Furthermore, Chee notes that linear amplification of ligated probes may be used to "determining a relative amount of the first and second amplicons wherein the relative amounts of the amplicons is indicative of the relative amounts of the first and second target sequence in the initial population" (p. 327, paragraph 423). Furthermore, Chee generally teaches modifications to different methods of amplification, including using one or more cycle of amplification and/or asymmetric amplification to provide flexibility in assay design. As noted by Chee, "The invention is directed to a variety of multiplexing methods used to amplify and/or genotypea variety of sample simultaneously" (Abstract). The stated goal of Chee is shared with the area of the prior art addressed by Andersen, therefore one of ordinary skill in the art at the time the invention was made would have been motivated to incorporate the method(s) of

amplification taught by Chee to the method of multiplex amplification taught by Andersen to achieve more universal amplification with a reasonable expectation for success.

6. Claims 6-7 and 22-23 are rejected under 35 U.S.C. 103(a) as being unpatentable over Andersen as applied to claims 1, 4-5, 8-16, 19-21, 24-33 and 35 above, and further in view of van Gelder (PNAS, 1990, vol. 87, p. 1663-1667). Andersen teaches methods of multiplex amplification of template nucleic acids for gene expression analysis (Abstract).

Andersen teaches the limitations of claims 1-5, 8-21, 24-33 and 35 as recited in the 102 rejection stated above.

Regarding claims 6-7 and 22-23, Wiley does not teach that the RNA sample comprises amplified antisense RNA. Van Gelder teaches a method of antisene RNA amplification.

With regard to claim 6 and 22, van Gelder teaches an embodiment of claims 1 or 16, wherein said sample of RNA comprises amplified antisense RNA (aRNA) (Figure 1, where antisense or aRNA is generated from ds cDNA molecules).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to have applied the method of cDNA synthesis and synthesis of amplified antisense RNA taught by van Gelder to the method of multiplex amplification taught by Andersen. As taught by van Gelder, "aRNA utilizes the specificity of the T7 RNA polymerase promoter site to allow in vitro amplification of a heterogeneous, complex population of cDNA" (p. 1666-1667, bottom of page to top of next). Furthermore, van Gelder notes that "our results indicate that the spectrum of aRNA produced during amplification qualitatively reflects the

population of cDNA from which it is produced. Since relative amounts of individual sequences present in the cDNA approximate their relative abundances in the transcribed RNA population, the amount of specific RNA in an aRNA population should reflect its abundance in the original RNA population" (p. 1667, col. 2, top paragraph). Therefore, one of ordinary skill in the art at the time the invention was made would have been motivated to incorporate the method of cDNA and aRNA synthesis taught by van Gelder to achieve amplification which maintains the relative abundance of transcripts within the original RNA sample with a reasonable expectation for success.

7. Claims 7 and 23 are rejected under 35 U.S.C. 103(a) as being unpatentable over Andersen as applied to claims 1, 4-5, 8-16, 19-21, 24-33 and 35 above, and further in view of Rougeon and Mach (PNAS, 1976, vol. 73, no. 10, p. 3418-3422; Rougeon herein). Andersen teaches methods of multiplex amplification of template nucleic acids for gene expression analysis (Abstract).

Regarding claims 7 and 23, Andersen does not teach that first strand cDNA is synthesized through self-priming without the addition of exogenous primers. Rougeon teaches self-primed synthesis of first strand cDNA (Abstract).

With regard to claim 7 and 23, Rougeon teaches an embodiment of claim 1, wherein said first strand cDNA is synthesized through RNA self-priming without addition of any exogenous synthetic primers (Abstract).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to have applied the method of cDNA synthesis taught by Rougeon to the

method of multiplex amplification taught by Andersen. As taught by Rougeon, "cDNA without homopolymeric tails, was also efficiently copied in the absence of an oligonucleotide primer, by DNA polymerase of avian myeloblastosis virus or E. coli" (Abstract). One of ordinary skill in the art at the time the invention was made would have been motivated to incorporate the method of cDNA synthesis taught by Rougeon to synthesize cDNA without exogenous primers with a reasonable expectation for success in order to achieve the initial sample for gene expression analysis without the need for additional primers.

Conclusion

No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Stephanie K. Mummert, Ph.D. whose telephone number is 571-272-8503. The examiner can normally be reached on M-F, 8:30-5.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 571-272-0782. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

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